

FORM PTO-1390 (Modified)  
(REV 11-98)

U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE

ATTORNEY'S DOCKET NUMBER

**TRANSMITTAL LETTER TO THE UNITED STATES  
DESIGNATED/ELECTED OFFICE (DO/EO/US)  
CONCERNING A FILING UNDER 35 U.S.C. 371**

PF 103 PCT US

U.S. APPLICATION NO. (IF KNOWN, SEE 37 CFR

**09/937673**INTERNATIONAL APPLICATION NO.  
**FR00/00822**INTERNATIONAL FILING DATE  
**March 31, 2000**PRIORITY DATE CLAIMED  
**March 31, 1999**

## TITLE OF INVENTION

**Bioprecursors of a retinoic derivative and pharmaceutical and/or cosmetic compositions**

## APPLICANT(S) FOR DO/EO/US

**Daniel Redoules, Roger Tarrous, Didier Fournier and Jean-Jacques Perie**

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

1. ☒ This is a **FIRST** submission of items concerning a filing under 35 U.S.C. 371.
2. ☐ This is a **SECOND** or **SUBSEQUENT** submission of items concerning a filing under 35 U.S.C. 371.
3. ☐ This is an express request to begin national examination procedures (35 U.S.C. 371(f)) at any time rather than delay examination until the expiration of the applicable time limit set in 35 U.S.C. 371(b) and PCT Articles 22 and 39(1).
4. ☒ A proper Demand for International Preliminary Examination was made by the 19th month from the earliest claimed priority date.
5. ☒ A copy of the International Application as filed (35 U.S.C. 371 (c) (2))
  - a. ☐ is transmitted herewith (required only if not transmitted by the International Bureau).
  - b. ☒ has been transmitted by the International Bureau.
  - c. ☐ is not required, as the application was filed in the United States Receiving Office (RO/US).
6. ☒ A translation of the International Application into English (35 U.S.C. 371(c)(2)).
7. ☒ A copy of the International Search Report (PCT/ISA/210).
8. ☐ Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371 (c)(3))
  - a. ☐ are transmitted herewith (required only if not transmitted by the International Bureau).
  - b. ☐ have been transmitted by the International Bureau.
  - c. ☐ have not been made; however, the time limit for making such amendments has NOT expired.
  - d. ☐ have not been made and will not be made.
9. ☐ A translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
10. ☒ An oath or declaration of the inventor(s) (35 U.S.C. 371 (c)(4)).
11. ☒ A copy of the International Preliminary Examination Report (PCT/IPEA/409).
12. ☐ A translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371 (c)(5)).

**Items 13 to 20 below concern document(s) or information included:**

13. ☐ An Information Disclosure Statement under 37 CFR 1.97 and 1.98.
14. ☐ An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
15. ☒ A **FIRST** preliminary amendment.
16. ☐ A **SECOND** or **SUBSEQUENT** preliminary amendment.
17. ☐ A substitute specification.
18. ☐ A change of power of attorney and/or address letter.
19. ☒ Certificate of Mailing by Express Mail
20. ☐ Other items or information:

U.S. APPLICATION NO. (IF KNOWN, SEE 37 CFR 1.53) <div style="font-size: 2em; font-weight: bold; margin-top: 5px;">09/937673</div>		INTERNATIONAL APPLICATION NO. <div style="font-weight: bold; margin-top: 5px;">FR00/00822</div>		ATTORNEY'S DOCKET NUMBER <div style="font-weight: bold; margin-top: 5px;">PF 103 PCT US</div>	
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21. The following fees are submitted: <b>BASIC NATIONAL FEE ( 37 CFR 1.492 (a) (1) - (5) ) :</b> <div style="margin-top: 5px;"> <input type="checkbox"/> Neither international preliminary examination fee (37 CFR 1.482) nor international search fee (37 CFR 1.445(a)(2)) paid to USPTO and International Search Report not prepared by the EPO or JPO ..... <span style="float: right;">\$1,000.00</span> </div> <div style="margin-top: 5px;"> <input checked="" type="checkbox"/> International preliminary examination fee (37 CFR 1.482) not paid to USPTO but International Search Report prepared by the EPO or JPO ..... <span style="float: right;">\$860.00</span> </div> <div style="margin-top: 5px;"> <input type="checkbox"/> International preliminary examination fee (37 CFR 1.482) not paid to USPTO but international search fee (37 CFR 1.445(a)(2)) paid to USPTO ..... <span style="float: right;">\$710.00</span> </div> <div style="margin-top: 5px;"> <input type="checkbox"/> International preliminary examination fee paid to USPTO (37 CFR 1.482) but all claims did not satisfy provisions of PCT Article 33(1)-(4) ..... <span style="float: right;">\$690.00</span> </div> <div style="margin-top: 5px;"> <input type="checkbox"/> International preliminary examination fee paid to USPTO (37 CFR 1.482) and all claims satisfied provisions of PCT Article 33(1)-(4) ..... <span style="float: right;">\$100.00</span> </div> <div style="text-align: right; margin-top: 10px;"> <b>ENTER APPROPRIATE BASIC FEE AMOUNT =</b> </div>				<b>CALCULATIONS PTO USE ONLY</b> <div style="border: 1px solid black; height: 100px; margin-top: 5px;"></div>	
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Surcharge of <b>\$130.00</b> for furnishing the oath or declaration later than <input type="checkbox"/> 20 <input type="checkbox"/> 30 months from the earliest claimed priority date (37 CFR 1.492 (e)).				<b>\$860.00</b>	
				<b>\$0.00</b>	

CLAIMS	NUMBER FILED	NUMBER EXTRA	RATE		
Total claims	15 - 20 =	0	x \$18.00		\$0.00
Independent claims	1 - 3 =	0	x \$80.00		\$0.00
Multiple Dependent Claims (check if applicable). <input type="checkbox"/>					\$0.00
<b>TOTAL OF ABOVE CALCULATIONS =</b>					<b>\$860.00</b>

Reduction of 1/2 for filing by small entity, if applicable. Verified Small Entity Statement must also be filed (Note 37 CFR 1.9, 1.27, 1.28) (check if applicable). <input type="checkbox"/>				<b>\$0.00</b>	
<b>SUBTOTAL =</b>					<b>\$860.00</b>

Processing fee of <b>\$130.00</b> for furnishing the English translation later than <input type="checkbox"/> 20 <input type="checkbox"/> 30 months from the earliest claimed priority date (37 CFR 1.492 (f)).				<b>\$0.00</b>	
<b>TOTAL NATIONAL FEE =</b>					<b>\$860.00</b>

Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31) (check if applicable). <input type="checkbox"/>				<b>\$0.00</b>	
<b>TOTAL FEES ENCLOSED =</b>					<b>\$860.00</b>

				Amount to be refunded \$	
				charged \$	

☒ A check in the amount of **\$860.00** to cover the above fees is enclosed.

☐ Please charge my Deposit Account No. \_\_\_\_\_ in the amount of \_\_\_\_\_ to cover the above fees.  
 A duplicate copy of this sheet is enclosed.


☒ The Commissioner is hereby authorized to charge any fees which may be required, or credit any overpayment to Deposit Account No. **8-3220** A duplicate copy of this sheet is enclosed.

**NOTE: Where an appropriate time limit under 37 CFR 1.494 or 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the application to pending status.**


SEND ALL CORRESPONDENCE TO:

**G. PATRICK SAGE**  
**THE FIRM OF HUESCHEN AND SAGE**  
**500 Columbia Plaza**  
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**Kalamazoo, MI 49007**

25666

  
PATENT TRADEMARK OFFICE

  
 SIGNATURE

**G. Patrick Sage**  
 NAME

**37,710**  
 REGISTRATION NUMBER

**September 28, 2001**  
 DATE

PF 103 PCT US/lw

\* \* \* \* \*

Applicants : Daniel Redoules, Roger Tarroux, Didier Fournier and  
Jean-Jacques Perie  
Title : Bioprecursors of a retinoic derivative and pharmaceutical and/or  
cosmetic compositions

\* \* \* \* \*

Honorable Commissioner of Patents and Trademarks  
Washington, D.C. 20231

PRELIMINARY AMENDMENT

Sir:

As soon as the Serial No. and Filing Date have been accorded the above-  
identified application, kindly enter the following amendment:

IN THE CLAIMS: Kindly cancel claims 1-15 and replace with the following claims 16-  
30, which correspond to each cancelled claim.

\*\*\*\*\*

REMARKS:

A few constructive editorial changes have been made in the claims to bring them  
somewhat more into line with U.S. practice and requirements.

Applicants have cancelled all of the originally filed claims, 1-15. New claims 16-  
30 have been added to better encompass the full scope and breadth of the invention,  
notwithstanding Applicants' belief that the claims would have been allowable as  
originally filed. Accordingly, Applicants assert that no claims have been narrowed within  
the meaning of Festo. The replacement Claims are attached hereto.

Entry of the amendments and favorable action on the merits are all hereby  
respectfully solicited.

Respectfully submitted,

THE FIRM OF HUESCHEN AND SAGE

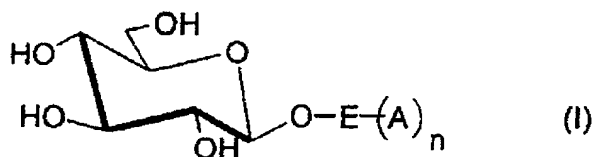
  
G. PATRICK SAGE, Attorney #37,710

Dated: September 28, 2001  
Customer No. 25,666  
500 Columbia Plaza  
350 East Michigan Ave.  
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(616) 382-0030

Enclosure: Return Postal Card Receipt  
Replacement Claims 16-30

## CLAIMS

16. A ternary glucosyl complex, which is a bioprecursor of at least one retinoic active principle, intended for percutaneous application, of formula (I)

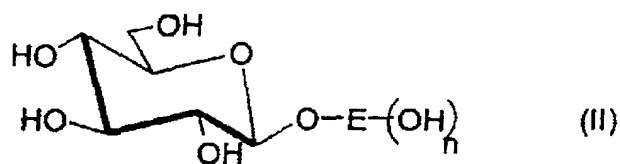


in which:

- E represents a linear, branched or cyclized hydrocarbon-based spacer group of aliphatic or aromatic nature which may contain one or more oxygen or hetero atoms and which may bear one or more carbonyl groups,
  - A represents a residue of a molecule of the retinoic active principle, linked to the spacer group via a carboxylate function,
  - $n = 1$  or  $2$ .
17. The glucosyl complex claim 16, wherein the retinoic active principle is retinoic acid.
18. The glucosyl complex of claim 16, wherein the group E represents a group which has a complementary pharmaceutical and/or cosmetic activity.
19. The glucosyl complex of claim 16, wherein the group E has a moisturizing, depigmenting and/or antibacterial activity.
20. The glucosyl complex of claim 16, wherein the group E represents a group derived from L or D glycerol, hydroquinone or flavonoids, in particular flavonoids of natural origin.
21. The glucosyl complex of claim 16, which is selected from:
- para-retinoyl-phenyl-glucopyranoside,
  - directinoyl-1,2-propanyl-glucopyranoside,
  - daidzin retinoate, and
  - genistin retinoate.
22. A pharmaceutical or cosmetic composition for topical use, which contains a glucosyl complex of claim 16, combined with a vehicle which is suitable for percutaneous administration.
23. The composition of claim 22, wherein, when it is applied to the skin, the complex undergoes an enzymatic double reaction, first of  $\beta$ -glucocerebrosidase type leading to hydrolysis between the glucose and the spacer group, and then of esterase type leading to hydrolysis between the spacer group and the active

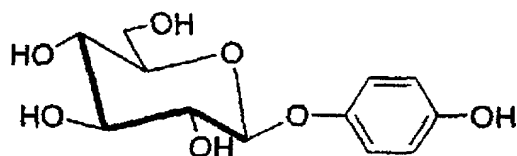
principle, the active principle thus being released in a delayed manner without an accumulation effect in the various layers of the skin.

24. The composition of claim 22, which contains from 0.001% to 10% by weight and preferably 0.01% to 0.1% by weight of glucosyl complex relative to the total weight of the composition.
25. The composition of claim 22, which is in the form of an emulsion.
26. The composition of claim 22, which is in the form of spherules, for instance liposomes, nanocapsules or nanospheres.
27. A process for preparing a complex of claim 16, wherein a compound of formula (II)

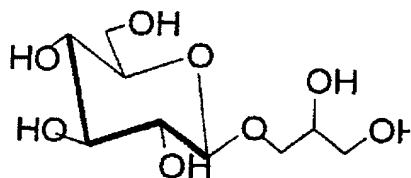


is reacted with the active principle in acid chloride form.

28. The process of claim 27, wherein the compound of formula II corresponds to formula IIa below:



29. The process of claim 27, wherein the compound of formula II corresponds to formula IIb below:



30. The process of claim 27, wherein the acid chloride is retinoyl chloride.

WO 00/58325

PCT/FR00/00822

**BIOPRECURSORS OF A RETINOIC DERIVATIVE AND  
PHARMACEUTICAL AND/OR COSMETIC COMPOSITIONS**

The present invention relates to a cosmetic or  
5 pharmaceutical composition for application to the skin,  
containing a compound capable of releasing two active  
substances by action of two enzymatic activities, the  
activities of glucocerebrosidase and esterase, starting  
with a glucoconjugate.

10

It has been confirmed after overexpression of cutaneous  
 $\beta$ -glucocerebrosidase that this enzyme is indeed capable  
of recognizing and hydrolyzing such glucoconjugates,  
thus allowing a slow release of the active substance,  
15 without an accumulation effect.

The bioprecursor strategy has been used previously for  
the release of active agents in two previous cases:

- 20 - release of retinol from its ester with palmitic  
acid under the action of the esterase activity of  
the skin (J. Boenlein, et al. Characterization of  
esterase and alcohol dehydrogenase activity in  
skin. Metabolism of retinyl palmitate to retinol  
25 (Vitamin A) during percutaneous absorption. Pharm.  
Res. 11, 1155-1159 (1994);
- release of vitamin C from a glucoconjugate under  
the action in this case of a glucosidase activity  
(patent FR-2 715 565).

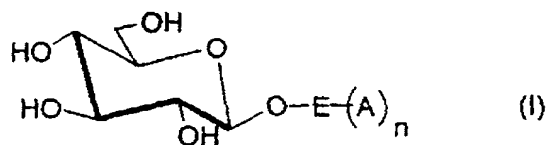
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Retinoic derivatives are nowadays used in dermatology  
in various indications, for instance psoriasis or  
ichthyosis, or alternatively to obtain a depigmentation  
of the skin (reduction in melanogenesis due to the  
35 action of vitamin A); applications to combat ageing of  
the skin are also sought.

However, the topical use of retinoic derivatives comes up against a certain number of difficulties, due to the lack of stability over time and the lack of stability to light of these derivatives, the irritation resulting from local overconcentrations and also from a poor penetration of these derivatives through the horny layer. This drawback is due to the highly lipophilic nature of the substance which, when deposited on the skin, is in fact largely removed by desquamation. Moreover, side effects (appearance of redness, irritation, oedema and excessive desquamation) limit its use to patients in urgent need, such as those afflicted with persistent acne.

This consequently explains the advantage of the present invention for improving the bioavailability of the active agent in the form of a glucose-spacer-active agent ternary complex, with facilitated penetration and hence the ability to be used in small amount, thus avoiding the harmful effects of local overconcentrations, which are responsible for intolerances.

The present invention relates to a ternary glucosyl complex, which is a bioprecursor of at least one retinoic active principle, in particular retinoic acid, intended for percutaneous application, of formula (I)



30

in which:

- E represents a linear, branched or cyclized hydrocarbon-based spacer group of aliphatic or aromatic nature which may contain one or more

oxygen hetero atoms and which may bear one or more carbonyl groups,

- A represents a residue of a molecule of said retinoic active principle, linked to the spacer group via a carboxylate function,
- n = 1 or 2.

According to another characteristic of the invention, in the complex of formula I, the group E represents a group which has a complementary pharmaceutical and/or cosmetic activity, in particular which has a moisturizing, depigmenting and/or antibacterial activity.

In particular, the group E can represent a group derived from L or D glycerol, hydroquinone or flavonoids, in particular flavonoids of natural origin.

As specific examples of glucosyl complexes according to the invention, mention will be made of:

- para-retinoyl-phenyl-glucopyranoside,
- diretinoyl-1,2-propanyl-glucopyranoside,
- daidzin retinoate, and
- genistin retinoate.

The present invention also covers pharmaceutical or cosmetic compositions for topical use, containing a glucosyl complex as defined above, combined with a vehicle which is suitable for percutaneous administration.

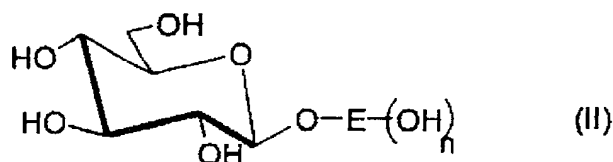
In accordance with the present invention, when said composition is applied to the skin, the complex undergoes an enzymatic double hydrolysis, first of  $\beta$ -glucocerebrosidase type leading to hydrolysis between the glucose and the spacer group, and then of esterase type leading to hydrolysis between the spacer group and the active principle, said active principle thus being



released in a delayed manner without an accumulation effect in the various layers of the skin.

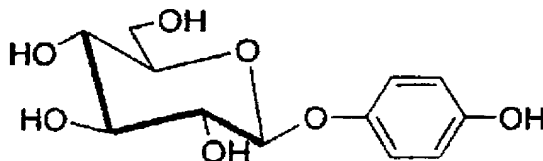
Advantageously, the composition according to the invention contains from 0.001% to 10% by weight and preferably from 0.01% to 0.1% by weight of glucosyl complex relative to the total weight of the composition.

The present invention also covers a process for preparing the glucosyl complexes defined above, which is characterized in that a compound of formula II

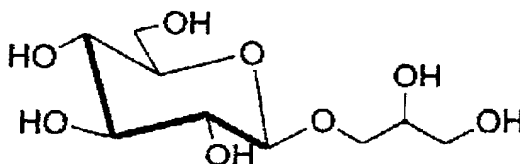


is reacted with the active principle in acid chloride form.

According to another characteristic of the invention, the compound of formula II corresponds to the more specific formula IIa below



According to another characteristic, the compound of formula II corresponding to formula IIb below



Finally, according to a last characteristic of the invention, the process involves reaction between the compounds of formula II, IIa or IIb with retinoyl chloride.

5

The glucose-spacer-active agent complex, after rapid migration into the first layers of the epidermis on account of its amphiphilic nature, is recognized as a pseudosubstrate by the two enzymatic activities  
10 involved:  $\beta$ -glucocerebrosidase (EC 3.2.1.45) responsible for the hydrolysis between glucose and spacer, and then esterase responsible for the second hydrolysis between spacer and active agent. Needless to say, the spacer may itself be chosen as active agent:  
15 this is achieved herein by using hydroquinone as spacer, which is itself active as a depigmenting or antibacterial agent. Two conjugate effects are thus obtained with a single formulation.

20 It has been demonstrated that the glucoconjugates described in the invention allow a genuine stabilization of retinoic active agents and also very good penetration: whereas derivatives that are too lipophilic, for instance retinoic acid or vitamin E  
25 ( $\alpha$ -tocopherol), accumulate in the upper layers of the stratum corneum after topical application and are removed by desquamation, their glucoconjugates, on the contrary, included in the same excipient, are partly found (portion not yet hydrolyzed) in the upper layers  
30 and also in the lower layers of the stratum corneum, for several days after they have been applied.

The design of these glucoconjugates as pseudosubstrates directed toward the  $\beta$ -glucocerebrosidase activity for  
35 the first hydrolysis is justified by several factors:

- this enzyme is accessible from the skin surface, as has been shown by topical application of a

specific inhibitor (W. M; Holleran, P. M Elias. J. Lipid. Res. 1994, 35. 905);

- this enzymatic activity which is predominant in the formation of the lipids of the skin surface (40% of the lipids result from this activity) is well conserved firstly between individuals and secondly in the course of the cycle of the seasons;
- under the conditions used in the present invention, this activity is sufficient since it is greater than the esterase activity (example 1).

This enzyme has thus been overexpressed; this has made it possible to determine the kinetic parameters of the substrates relative to a reference. Values are given by way of example for two conjugates, one containing two components and the other containing three components. The values indicate that these pseudosubstrates are better recognized than the reference substrate ( $K_M$  values), which is explained by the more lipophilic nature of these conjugates relative to the reference 4-methylumbelliferyl-glucopyranoside with respect to an enzyme whose substrate is itself highly lipophilic ( $\beta$ -glucosyl-ceramide); moreover, the  $V_m$  values show that the active agents are indeed released, with kinetics that are compatible with the intended objective, namely an effect over time starting with a pseudosubstrate applied to the skin in minimal amount but which will be integrally used.

The strategy presented above may be extended and modified in different directions. By way of example:

*- modification of the spacer:*

The spacer may be modified into a structure which is closer to that of the natural substrate ( $\beta$ -glucocerebroside) in which the spacer is related to glycerol. The corresponding glucose-glycerol (L or D)-

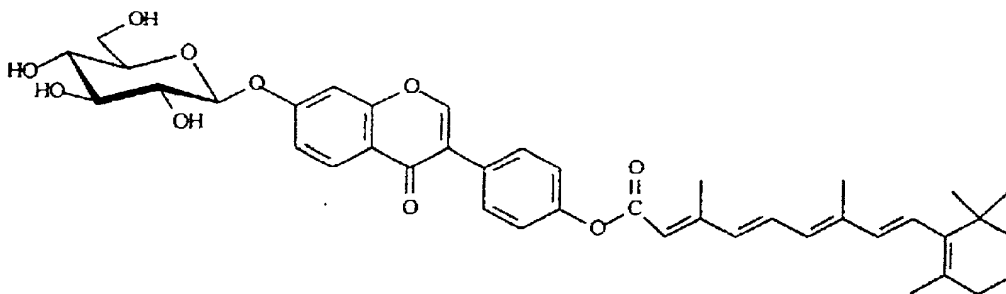
retinoic acid glucoconjugate has also been synthesized and studied. It should be noted that the two free hydroxyl groups on the glycerol allow the attachment in ester form of two retinoic units per complex molecule.

5 In this case, the action complementary to the retinoic activity is that of a moisturizing effect provided by in situ release of glycerol;

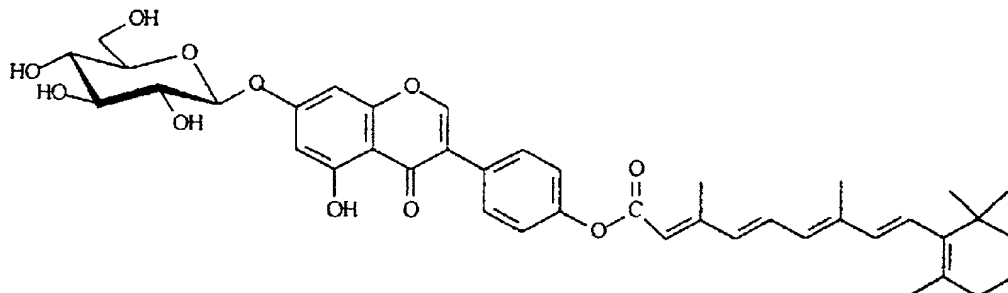
- combination with the retinoic activity of antioxidant  
10 properties of flavonoids:

A certain number of flavonoids of natural origin are combined with a saccharide portion which gives them amphiphilic properties.

15 This is the case, for example, for genistins or daidzin. The absorption of such compounds by the skin surface is thus assured. This first antioxidant activity is combined with the retinoic activity by  
20 attachment of one or more retinoic acid molecules per flavone unit. The corresponding structures are given below:



25 daidzin retinoate



genistin retinoate

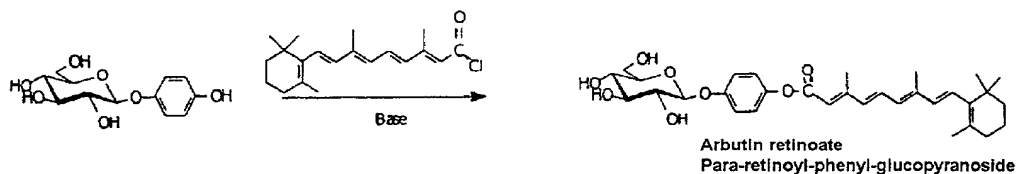
5 In conclusion, the present invention shows the part which may be taken from the  $\beta$ -glucocerebrosidase and esterase activities of the skin surface to obtain the release of various types of active agents from glucosyl bioprecursors.

10 The structure of the corresponding glucoconjugates ensures good penetration on account of their amphiphilic nature and thus optimal use, very good recognition by the first enzyme,  $\beta$ -glucocerebrosidase,  
15 on account of the presence of one or more lipophilic retinyl residues, and a release of the active agents with kinetics which ensure effective cleavage and an effect with remanence over time.

20 The syntheses of the glucoconjugates, their formulation and their activity as pseudosubstrates are described below:

a) Synthesis of the bioprecursors

25 Arbutin retinoate (p-retinoyl-phenyl-glucopyranoside) is prepared from arbutin and retinoyl chloride according to the following reaction scheme.

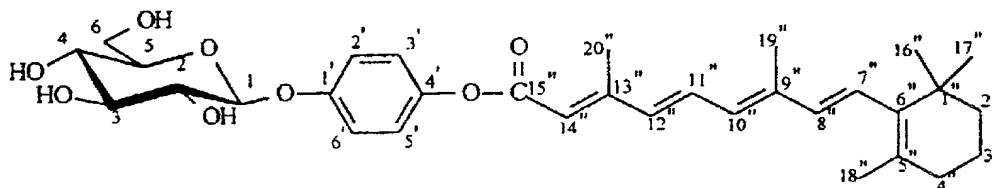


This coupling reaction results from a selective and  
 5 initial dehydrogenation of the phenol function followed  
 by a nucleophilic attack of the phenoxy formed on the  
 acid chloride. The selective dehydrogenation is  
 obtained by the addition, to the maximum, of one  
 equivalent of base (generally 0.9 equivalent) reacting  
 10 with the phenol group, the pKa value of which (pKa = 9)  
 is much lower than that of the other hydroxyl functions  
 of the glucose portion (pKa > 16).

#### Preparation of retinoyl chloride

15 0.41 g (3.3 mmol) of thionyl chloride in methylene  
 chloride (2 ml) is added dropwise to a suspension of  
 1 g (3.32 mmol) of retinoic acid in 15 ml of anhydrous  
 methylene chloride cooled to 0°C, maintained under  
 20 argon and containing 0.32 g of pyridine (0.4 mmol). The  
 mixture is allowed to warm to room temperature and  
 stirring is continued for one hour. The red syrup  
 obtained is filtered through glass wool and is  
 immediately used in the following step.

25 Preparation of arbutin retinoate (p-retinoyl-phenyl-  
 glucopyranoside)



30 0.7 g (2.6 mmol) of arbutin is added dropwise to a  
 suspension of 50 mg (2.1 mmol) of sodium hydride in

10 ml of anhydrous DMF cooled to 0°C and maintained under argon. The 15 ml of the retinoyl chloride solution prepared above are added slowly and the mixture is stirred for one hour while allowing it to  
5 warm to room temperature. The excess acid chloride is hydrolyzed with 5 ml of water and the mixture is neutralized by addition of a few drops of saturated sodium hydrogen carbonate solution. The organic phase extracted, dried and evaporated under vacuum is  
10 purified by HPLC (C18: MeOH-H<sub>2</sub>O: 85-15).

1.1 g of red crystals are obtained. Yield = 73%.

<sup>1</sup>H NMR (CDCl<sub>3</sub>) δ ppm:

15

6.9-7.1 (m, 5H, H-2', 3', 5', 6', 11''), 6.1-6.35 (m, 4H, H-7'', 8'' -CH=CH, 10'', 12'' -CH=CH), 5.88 (s, 1H, H-14'' -CH=CH-), 4.84 (d, 1H, H-1), 3.3-3.9 (m, 6H, H-2, 3, 4, 2 H6), 2.3 (1 s, 3H, H-20'' -CH<sub>3</sub>), 1.97-2.03 (1s  
20 and m, 5H, H-19'' -CH<sub>3</sub>, 4'' -CH<sub>2</sub>), 1.36-1.68 (1m, 1s, 7H, H-2'', 3'' -(CH<sub>2</sub>)<sub>2</sub>, 18'' -CH<sub>3</sub>), 1.02 (1s, 6H, H-16'', 17'' -Cme<sub>2</sub>).

<sup>13</sup>C NMR (CDCl<sub>3</sub>) δ ppm:

25

166 (C-15''), 155.5 (C-13''), 154.7 (C-1'), 145.6 (C-4'), 140.2 (C-9''), 137.7 (C-6''), 137.4 (C-8''), 135.1 (C-12''), 131.9 (C-11''), 130 (C-5''), 129.7 and 128.9 (C-10'', 7''), 122.8 (C-3', 5'), 117.8 (C-14''), 117.4 (C-2', 6'), 100.1 (C-1), 75.7 (C-3), 75 (C-5), 71.5  
30 (C-2), 70.1 (C-4), 61.7 (C-6), 39.6 (C-2''), 34.3 (C-1''), 33.2 (C-4''), 29 (C-16'', 17''), 21.8 (C-18''), 19.3 (C-3''), 14.1 and 13 (C-20'', 19'')

IR: 3418 cm<sup>-1</sup> OH, 1700 cm<sup>-1</sup> (ester C=O), 1684, 1576,  
35 1504, 1447, 1358, 1195, 1129 cm<sup>-1</sup> (CO)

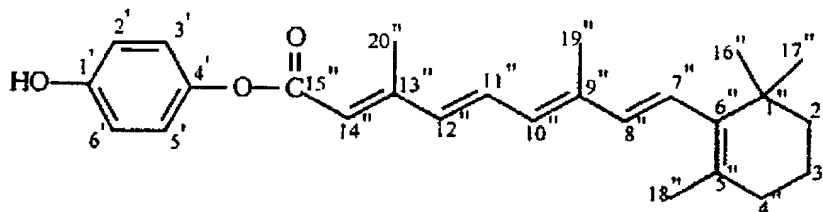
MS (m/z) 555 (M<sup>+</sup>+1), 577 (M<sup>+</sup>+Na).

With the aim of studying the kinetics of cleavage of arbutin retinoate by β-glucocerebrosidase, we

synthesized its hydrolysis product: 4-hydroxyphenyl p-retinoate.

# Preparation of phenol p-retinoate

5



300 mg of dried  $\text{Na}_2\text{CO}_3$  (2.8 mmol) are added to a solution of hydroquinone (300 mg, 2.7 mmol) in anhydrous acetone (15 ml) maintained under argon, followed by slow addition of the 15 ml of retinoyl chloride solution (max 3 mmol) prepared above. After stirring for one hour, the excess acid chloride is hydrolyzed by adding 5 ml of water and the medium is neutralized by adding a few drops of saturated  $\text{NaHCO}_3$  solution. The organic phase extracted, dried, evaporated under vacuum and purified by HPLC ( $\text{C}_{18}$ : eluant  $\text{MeOH-H}_2\text{O}$ : 90-10) gives 0.61 g of red crystals (yield = 52%).

20

$^1\text{H}$  NMR ( $\text{CDCl}_3$ )  $\delta$  ppm:

6.95 and 6.78 (2d, 4H, H-2', 3', 5', 6', J - 11 Hz), 7.07 (dd, 1H, H-11''), 6.1-6.4 (m, 4H, H-7'', 8'' -CH=CH, 10'', 12'' -CH=CH), 5.8 (s, 1H, H-14'' -CH=CH-), 2.4 (1 s, 3H, H-20'' -CH<sub>3</sub>), 2-2.1 (1s and m, 5H, H-19'' -CH<sub>3</sub>, 4'' -CH<sub>2</sub>), 1.4-1.72 (1m, 1s, 7H, H-2'', 3'' -(CH<sub>2</sub>)<sub>2</sub>, 18'' -CH<sub>3</sub>), 1.02 (1s, 6H, H-16'', 17'' -Cme<sub>2</sub>).

$^{13}\text{C}$  NMR ( $\text{CDCl}_3$ )  $\delta$  ppm:

166.4 (C-15''), 155.3 (C-13''), 153.8 (C-1'), 143.9 (C-4'), 140.3 (C-9''), 137.7 (C-6''), 137.4 (C-8''), 135 (C-12''), 131.9 (C-11''), 130.2 (C-5''), 129.5 and 128.9 (C-10'', 7''), 122.8 (C-3', 5'), 117.8 (C-14''), 117.3 (C-2', 6'), 39.6 (C-2''), 34.3 (C-1''), 33.2 (C-4''), 29



(C-16", 17"), 21.8 (C-18"), 19.3 (C-3"), 14.2 and 13 (C-20", 19")

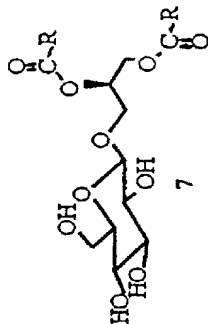
MS (FAB/MNBA) m/Z: 415 ( $M^+$ +Na).

5

Synthesis of the derivative diretinoyl-1,2-propanyl-glucopyranoside (glucose-glycerol-retinoic acid conjugate)

- 10 The figure below describes the reaction scheme we employed to carry out the synthesis of compounds **6** and **7** ( $C_2$  enantiomers of the glycerol spacer).

Figure 1



The selective deacylation in position 1 is obtained by aminolysis of the peracetyl glucopyranose 1 using ammonia in the mixture (THF-MeOH: 7-3).

5 The glucoconjugate 3 was prepared according to the Schmidt method (Schmidt, R.R. Angew. Chem. Int. Ed. Engl. 1986, 25, 212) which allows a stereoselective coupling using the imidate as nucleofugal activator.

10 This intermediate is synthesized by action of sodium hydride on the glucopyranose deprotected at C-1, which, when converted into an alkoxide, reacts as a nucleophile with trichloroacetonitrile to give the  $\alpha$ -imidate 2.

15 The IR spectrum of this compound shows the characteristic band at  $1670\text{ cm}^{-1}$  which may be attributed to the imine bond  $\text{C}=\text{N}$ . The  $^1\text{H}$  NMR spectrum of this compound has a doublet at 6.6 ppm which reflects the presence of the hydrogen at 1 coupled to the hydrogen on carbon C-2, in an  $\alpha$  configuration ( $J = 3.5\text{ Hz}$ ).

20 In the presence of Lewis acid ( $\text{BF}_3$  etherate), the tetraacetyl  $\alpha$ -imidate 2 reacts with an alcohol in methylene chloride and leads to the formation of the corresponding glucoconjugate. This reaction results from an initial activation of the imidate function with the Lewis acid, followed by a nucleophilic attack of the alcohol on carbon 1 of the saccharide portion to give the  $\beta$ -glucosyl derivative exclusively ( $J = 8\text{ Hz}$  at  $\text{C}_1$ ).

25 Deprotection of the tetraacetyl glucoconjugates is obtained by treatment with ion-exchange resin (Amberlyst A-26 (OH)) according to a series of ion exchanges at the surface of the resin.

A rapid filtration, after leaving in contact with the resin overnight, allows the deprotected water-soluble compound to be isolated readily in a good yield.

5 Silylation of the saccharide derivatives with TBDMS  
triflate generally gives only very poor yields  
(T. Limori, H. Takashashi and S. Ikegami, Tetrahedron  
Lett., 1996, 37, 649); we have developed conditions for  
10 of the glucopyranose. The structure of the derivative  
obtained is established by the  $^1\text{H}$  NMR spectra: the  
presence of the methyl protons of the TBDMS groups and  
their integration unequivocally establishes the  
tetrasilylation.

15 Selective hydrolysis of the acetal 4 without the  
concomitant loss of the silyl protections was able to  
be obtained in a yield of 66% using an excess of  
ethanedithiol in the presence of a catalytic amount of  
20 p-toluenesulfonic acid in methylene chloride. The  
structure of compound 5 is deduced from the IR spectrum  
(OH band at  $3390\text{ cm}^{-1}$ ), the mass spectrum (FAB  $\text{M}^+ + \text{Na} = 733$ )  
and the proton and  $^{13}\text{C}$  NMR spectra which show the  
disappearance of the methyls of the acetal.

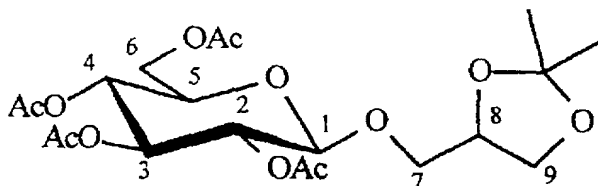
25 The double esterification is obtained in a yield of  
76%, according to the method applied above. Thus, in  
the presence of two equivalents of sodium hydride, the  
diol reacts with retinoyl chloride to give the expected  
30 diester. The spectral characteristics are in accordance  
with the proposed structure. The  $^1\text{H}$  and  $^{13}\text{C}$  NMR spectra  
show the presence of retinoic synthons and tetrasilyl  
glucose.

35 The final step of deprotection of the hydroxyl groups  
borne by the saccharide unit was then performed in  
anhydrous THF, in the presence of 4 equivalents of TBAF  
and gave the glucose-glycerol-retinoic acid conjugate 6  
in a yield of 90%.

(TBDMS = tert-butyldimethylsilyl; TBAF = tetra-n-butylammonium fluoride))

### Preparation of derivative 3

5



100 mg of  $\text{BF}_3$  etherate dissolved in 1 ml of  $\text{CH}_2\text{Cl}_2$  are added slowly to a mixture, cooled to  $-10^\circ\text{C}$ , of 1.6 g of imidate (4.6 mmol) and 0.6 g of  $\alpha,\beta$ -isopropylidene-glycerol (4.6 mmol) in 30 ml of  $\text{CH}_2\text{Cl}_2$ . Stirring is continued for two hours and the mixture is washed with saturated  $\text{NH}_4\text{Cl}$  and neutralized with saturated  $\text{NaHCO}_3$  solution. After drying ( $\text{MgSO}_4$ ), the resulting solution is concentrated under reduced pressure and the crude residue is purified by flash chromatography (eluent: hexane-ethyl acetate: 3-2). 1.74 g (3.8 mmol) of white crystals are obtained.

$^1\text{H}$  NMR  $\text{CDCl}_3$   $\delta$  ppm (300 MHz):

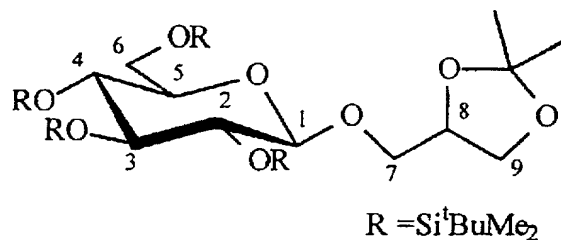
4.36-5.19 (m, 3H, H-1, 2, 3), 4.59 (dd, 1H, H-5), 4.23-3.57 (m, 8H, H-4, 8, 2H6, 2H7, 2H9), 1.96-2.07 (4s, 12H, Ac), 1.39 and 1.32 (2s, 6H,  $\text{CH}_3$  acetal).

$^{13}\text{C}$  NMR ( $\text{CDCl}_3$ )  $\delta$  ppm:

169.3-170.7 (4s, 4 OCOR), 109.4 ( $\text{C}_{\text{quat}}$ , isopropylidene), 101 (C-1), 74.2 (C-8), 72.8 (C-3), 71.9 (C-5), 71.2 (C-2), 69.2 (C-7), 68.4 (C-4), 66.8 (C-9), 61.9 (C-6), 26.6 and 25.4 ( $2\text{CH}_3$  of the acetals).

IR:  $1756\text{ cm}^{-1}$  (ester  $\text{C}=\text{O}$ ),  $1370$ ,  $1229$ ,  $1167$ ,  $1050\text{ cm}^{-1}$  (CO)

# Preparation of the silyl derivative 4



5

A solution of 400 mg (0.86 mmol) of the glucoconjugate 3 in 20 ml of MeOH containing 75 mg of Amberlyst A26 resin is stirred for 24 hours at ambient temperature. The filtered and concentrated solution gives 250 mg of deprotected glucopyranoside derivative (0.85 mmol).

1.8 g (6.8 mmol) of TBDMS triflate are added to a solution of the above deprotected derivative (250 mg) containing 1.1 g of lutidine (10 mmol) in 15 ml of anhydrous methylene chloride, cooled to 0°C and under argon. The mixture is stirred at ambient temperature for 30 hours. The organic solution washed, dried and evaporated under vacuum gives, after purification by flash chromatography, 0.4 g of colorless resin (eluent: hexane-EtOAc: 30-1).

$^1\text{H}$  NMR  $\text{CDCl}_3$   $\delta$  ppm (300 MHz):

4.68 (d, 1H, H-1,  $J_{\text{aa}}=10$  Hz), 4.32 (dd, 1H, H-3), 4.05 (t, 1H, H-8), 3.58-3.89 (m, 9H, H-2, 4, 5, 2H6, 2H7, 2H9), 1.35 and 1.41 (2s, 6H,  $\text{CH}_3$  acetal), 0.85-0.9 (4s, 36H, 4  $\text{Si}^t\text{Bu}$ ), 0.04-0.09 (4s, 24H,  $\text{SiMe}_2$ ).

$^{13}\text{C}$  NMR ( $\text{CDCl}_3$ )  $\delta$  ppm:

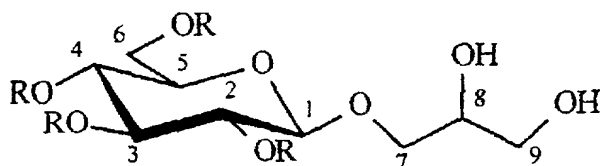
109 ( $\text{C}_{\text{quat}}$ , isopropylidene), 102.3 (C-1), 82.4 (C-3), 79.1 (C-5), 77.5 (C-2), 74.5 (C-8), 70.2 (C-4), 70.1 (C-7), 67.6 (C-9), 64.2 (C-6), 26.9 and 25.5 (2 $\text{CH}_3$  of the acetals), 25.9 ( $\text{CH}_3$  ( $^t\text{Bu}$ )), 17.9-18.4 (4s,  $\text{C}_{\text{quat}}\text{-Si}$ ), -4.11-(-5.4) (4s,  $\text{CH}_3\text{Si}$ ).

MS (FAB/ONPOE) m/z: 773 ( $M^+Na$ )

IR: 1472, 1361, 1255, 1096  $cm^{-1}$  (CO)

Preparation of the derivative 5

5



$R = Si^tBuMe_2$

0.88 g of ethanedithiol (9.33 mmol) and 25 mg of p-toluenesulfonic acid (0.132 mmol) are added, under argon and with mechanical stirring, to a solution of 1 g of 4 (1.33 mmol) in 20 ml of methylene chloride. Stirring is continued for a further 15 hours. After washing with saturated NaCl solution, drying ( $MgSO_4$ ) and then filtration, and after concentrating under vacuum, a residue is recovered which is purified by flash chromatography (hexane-ethyl acetate: 1-1). 0.625 g of colorless oil is thus collected (yield = 66%).

$^1H$  NMR  $CDCl_3$   $\delta$  ppm (300 MHz):  
4.67 (d, 1H, H-1, Jaa=10 Hz), 3.53-3.98 (m, 13H, H-2, 3, 4, 5, 2H6, 2H1', 2H2', 2H3', 2 OH), 0.85-0.9 (4s, 36H, 4  $Si^tBu$ ), 0.038-0.09 (4s, 24H,  $SiMe_2$ ).

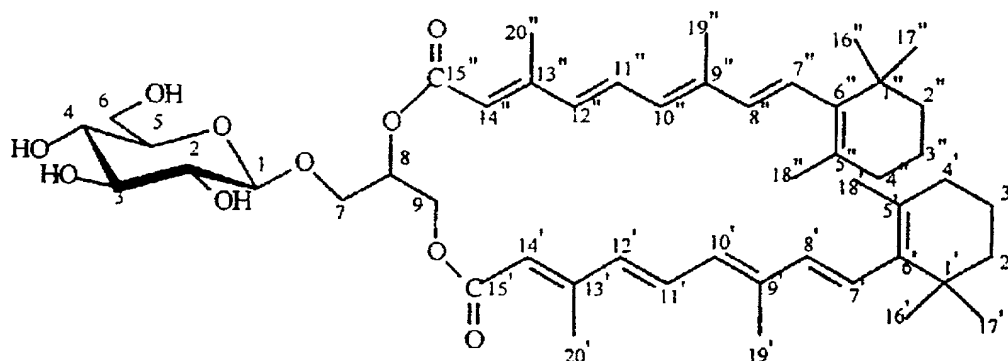
$^{13}C$  NMR ( $CDCl_3$ )  $\delta$  ppm:  
103.3 (C-1), 82.7 (C-3), 78.9 (C-5), 78.2 (C-2), 72.2 (C-1'), 71 (C-4), 70.2 (C-2'), 64.2 (C-6), 63.9 (C-3'), 25.9 ( $CH_3$  ( $^tBu$ )), 18.4-17.9 (4s,  $C_{quat}Si$ ), -4.11-(-5.4) (4s,  $CH_3Si$ ).

30

MS (FAB/ONPOE) m/z: 733 ( $M^+Na$ )

IR: 3390  $cm^{-1}$  (OH), 1384, 1218, 1078  $cm^{-1}$  (CO)

Preparation of the derivative 6 (S) or 7 (R)



5

The double esterification is carried out according to the procedure described for the synthesis of arbutin retinoate, but in this case we take methylene chloride as solvent and use 2 equivalents of sodium hydride. The separation of the esterified compound which is in the  $R_f$  zone = 0.2, eluted with the mixture (hexane-EtOAc: 25-1), is performed by flash chromatography.

The diester obtained (0.68 g, 0.53 mmol) is desilylated with 2.3 g of TBAF (7.4 mmol) in 15 ml of anhydrous THF. After stirring for 4 hours, washing the extract and evaporation to dryness, the product is purified by TLC on silica gel, type 60, in a  $\text{CH}_2\text{Cl}_2$ -MeOH mixture (95-5),  $R_f$  = 0.3. 0.4 g of red crystals is isolated.

20

(6,  $\alpha_D = -8^\circ$ , S form)

(7,  $\alpha_D = +12^\circ$ , R form)

$^1\text{H}$  NMR  $\text{CDCl}_3$   $\delta$  ppm (300 MHz):

6.97 (dd, 2H, H-11', 11''C=CH,  $J_{\text{H}}=16$  Hz), 6.08-6.3 (m, 8H, H-7', 7'', 8', 8''-HC=CH, H-10', 10'', 12', 12''-C=CH), 5.74 (s, 2H, H-14', 14''-CH=CH), 4.32-4.37 (m, 2H, H-1.8), 3.23-3.96 (m, 14H, H-2, 3, 4, 5, 2H6, 2H7, 2H9), 2.3 (s, 6H, H-20', 20''-CH<sub>3</sub>), 1.97-2.03 (1s and m, 10H, H-19', 19''-CH<sub>3</sub>, H-4', 4''-CH<sub>2</sub>), 1.36-1.68 (1m, 1s, 14H, H-2', 3', 2'', 3''-(CH<sub>2</sub>)<sub>2</sub>, H-18', 18''



-CH<sub>3</sub>), 0.94, 0.98, 1, 1.01, (4s, 12H, H-16', 16'', 17', 17'' -CMe<sub>2</sub>).

<sup>13</sup>C NMR (CDCl<sub>3</sub>) δ ppm:

5 167, 166.5 (C-15', 15''), 154.3, 153.8 (C-13', 13''), 140 (C-9', 9''), 137.7 (C-6', 6''), 137.3 (C-8, 8''), 135.1 (C-12', 12''), 131.6 (C-11', 11''), 130.4 (C-5', 5''), 129.6 and 128.8 (C-10', 10'', 7', 7''), 117.9 (C-14', 14''), 103.7 (C-1), 76.1 (C-8), 73.7 (C-3,5), 70  
10 (C-2,4), 68.3 (C-7), 62.5 (C-9), 62 (C-6), 39.6 (C-2', 2''), 34.3 (C-1', 1''), 33.2 (C-4', 4''), 29 (C-16', 16'', 17', 17''), 21.8 (C-18', 18''), 19.3 (C-3', 3''), 13.8 and 13 (C-20', 20'', 19', 19'')

IR: 3427 cm<sup>-1</sup> OH, 1706 cm<sup>-1</sup> (ester C=O), 1609, 1457,  
15 1384, 1237, 1141, 1083 cm<sup>-1</sup> (CO)

MS (FAB/MNBA) m/z: 841 (M<sup>+</sup>+Na)

#### b) Formulations

20 The compositions according to the invention contain from 0.001% to 10% by weight and preferably 0.01% to 0.1% by weight of active precursors relative to the total weight of the composition.

25 The composition according to the invention may be in the form of an oil-in-water (O/W) or water-in-oil (W/O) emulsion. It may also be in the form of spherules, for instance liposomes, nanocapsules or nanospheres.

30 When the composition is an emulsion, the proportion of the fatty phase ranges from 5% to 80% by weight and preferably from 5% to 50% by weight relative to the total weight of the composition. The oils, emulsifiers and coemulsifiers used in the composition, in emulsion  
35 form, are chosen from those conventionally used in cosmetics. The emulsifier and coemulsifier are present in the composition in a proportion ranging from 0.3% to 10% by weight relative to the total weight of the composition.

The composition according to the invention may also contain acceptable cosmetic or dermatological additives. These additives may be, in particular, antioxidants, bioprecursors of these antioxidants, for instance  $\delta$ -tocopherylglucopyranoside, surfactants, fatty substances, moisturizers, preserving agents, fragrances, gelling agents, chelating agents, pigments, for instance titanium oxide, screening agents and free vitamins, for instance ascorbic acid.

c) Enzymatic study

- Comparison of the  $\beta$ -glucocerebrosidase and esterase activities

The stripping technique allows these two different activities to be assayed accurately using the same sample. To do this, we used two artificial substrates, 4-methyl-umbelliferyl- $\beta$ -D-glucopyranoside (2 mM), to assay the  $\beta$ -glucocerebrosidase activity, and 4-methyl-umbelliferyl-palmitate (2 mM) for that of the esterases.

The following table gives the amount of 4-methyl-umbelliferone released after hydrolysis for one hour by the  $\beta$ -glucocerebrosidase and esterase extracted from three 25 cm<sup>2</sup> strips.

It is noted that, at skin pH (pH = 5.5), the  $\beta$ -glucocerebrosidase activity is on average twice as high as that of esterase.

	$\beta$ -glucocerebrosidase	Esterase
Weighted activities Nmol/hour/ $\mu$ g of total proteins	0.23 $\pm$ 0.1	0.13 $\pm$ 0.08

- Recognition and hydrolysis of the pseudosubstrates

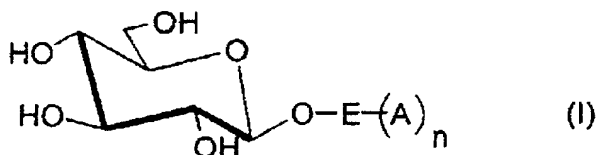
After confirming that  $\beta$ -glucocerebrosidase is expressed in the keratinocytes, we produced a recombinant enzyme in the baculovirus system. A histidine tail was added to the COOH end of the protein to allow it to be purified by affinity column chromatography.

Thus, we were able to determine the Michaelis constants ( $K_m$ ) and the  $V_m$  values of the recombinant  $\beta$ -glucocerebrosidase in particular for the retinoic acid-arbutin glucoconjugate. The kinetics measurements are carried out in a phthalate buffer at pH 5.6 (0.025 M) containing taurocholate (5 mM), purified  $\beta$ -glucocerebrosidase and the test conjugate at various concentrations. The incubation lasts 30 minutes and the amount of hydroquinone-retinoic acid conjugate released is assayed by HPLC. The table below gives the results obtained. It shows, in terms of affinity, that the two test glucoconjugates are much better substrates than the reference. As regards their rates of hydrolysis, these are lower and thus allow effects to be obtained over time.

Substrates	$K_m$	$V_m$ weighted by the amount of soluble proteins
4-Methylumbelliferyl-glucopyranoside	$2.8 \pm 0.7 \text{ mM}$	$4000 \pm 1000 \text{ nmol/h/mg}$
$\delta$ -Tocopheryl-glucopyranoside	$7 \pm 1 \text{ } \mu\text{M}$	$453 \pm 20 \text{ nmol/h/mg}$
Arbutin retinoate	$5 \pm 1.2 \text{ } \mu\text{M}$	$235 \pm 19 \text{ nmol/h/mg}$
Diretinyl-glyceryl-glucopyranoside (R)	$8.6 \pm 2.5 \text{ } \mu\text{M}$	$74 \pm 7 \text{ nmol/h/mg}$
Diretinyl-glycerol-glucopyranoside (S)	$5 \pm 0.4 \text{ } \mu\text{M}$	$17 \pm 0.4 \text{ nmol/h/mg}$

CLAIMS

1. A ternary glucosyl complex, which is a bioprecursor of at least one retinoic active principle, intended for percutaneous application, of formula (I)



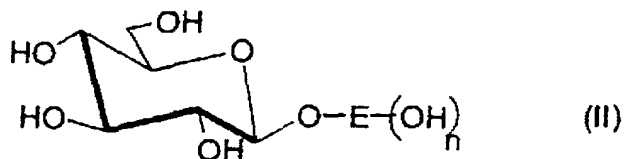
in which:

- E represents a linear, branched or cyclized hydrocarbon-based spacer group of aliphatic or aromatic nature which may contain one or more oxygen hetero atoms and which may bear one or more carbonyl groups,
- A represents a residue of a molecule of said retinoic active principle, linked to the spacer group via a carboxylate function,
- $n = 1$  or  $2$ .

2. The glucosyl complex as claimed in claim 1, characterized in that the retinoic active principle is retinoic acid.
3. The glucosyl complex as claimed in either of claims 1 and 2, characterized in that the group E represents a group which has a complementary pharmaceutical and/or cosmetic activity.
4. The glucosyl complex as claimed in one of claims 1 to 3, characterized in that the group E has a moisturizing, depigmenting and/or antibacterial activity.

5. The glucosyl complex as claimed in one of claims 1 to 4, characterized in that the group E represents a group derived from L or D glycerol, hydroquinone or flavonoids, in particular flavonoids of natural origin.
6. The glucosyl complex as claimed in one of claims 1 to 5, characterized in that it is chosen from:
- para-retinoyl-phenyl-glucopyranoside,
  - diretinoyl-1,2-propanyl-glucopyranoside,
  - daidzin retinoate, and
  - genistin retinoate.
7. A pharmaceutical or cosmetic composition for topical use, characterized in that it contains a glucosyl complex as claimed in one of claims 1 to 6, combined with a vehicle which is suitable for percutaneous administration.
8. The composition as claimed in claim 7, characterized in that, when it is applied to the skin, said complex undergoes an enzymatic double reaction, first of  $\beta$ -glucocerebrosidase type leading to hydrolysis between the glucose and the spacer group, and then of esterase type leading to hydrolysis between the spacer group and the active principle, said active principle thus being released in a delayed manner without an accumulation effect in the various layers of the skin.
9. The composition as claimed in either of claims 7 and 8, characterized in that it contains from 0.001% to 10% by weight and preferably 0.01% to 0.1% by weight of glucosyl complex relative to the total weight of the composition.

10. The composition as claimed in one of claims 7 to 9, characterized in that it is in the form of an emulsion.
- 5 11. The composition as claimed in one of claims 7 to 9, characterized in that it is in the form of spherules, for instance liposomes, nanocapsules or nanospheres.
- 10 12. A process for preparing a complex as claimed in one of claims 1 to 6, characterized in that a compound of formula (II)

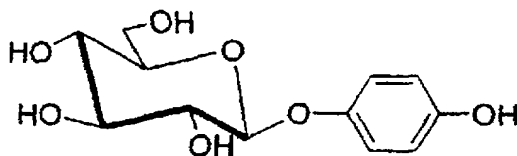


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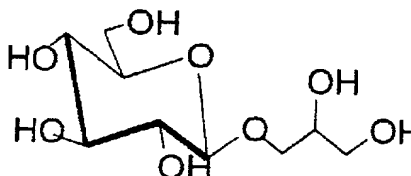
is reacted with the active principle in acid chloride form.

13. The process as claimed in claim 12, characterized in that the compound of formula II corresponds to formula IIa below:

20



- 25 14. The process as claimed in claim 12, characterized in that the compound of formula II corresponds to formula IIb below:



15. The process as claimed in one of claims 12 to 14, characterized in that the acid chloride is retinoyl chloride.

## DECLARATION AND POWER OF ATTORNEY FOR PATENT APPLICATION

As a below named inventor, I hereby declare that:

My residence, post office address and citizenship are as stated below, next to my name, and

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled **BIOPRECURSORS OF A RETINOIC DERIVATIVE AND PHARMACEUTICAL AND/OR COSMETIC COMPOSITIONS**

de la surface cutanée et compositions pharmaceutiques et/ou cosmétiques  
the specification of which (check one of the following)

is attached hereto

**X** was filed on MARCH 31, 2000 as **International**  
Application Serial No. PCT/FR00/00822  
And was amended on \_\_\_\_\_  
(if applicable)

I hereby state that I have reviewed and understand the contents of the above-identified specification, including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose information which is material to the examination of this application in accordance with Title 37, Code of Federal Regulations, §1.56(a).

I hereby claim foreign priority benefits under Title 35, United States Code, §119 of any foreign application(s) for patent or inventor's certificate listed below and have also identified below any foreign application for patent or inventor(s) certificate having a filing date before that of the application on which priority is claimed:

<u>Application Serial Number</u>	<u>Country</u>	<u>Filing Date (Day/Month/Year)</u>	<u>Priority Claimed (yes/no)</u>
99 04032	FRANCE	31/March/1999	Yes

I hereby claim the benefit under Title 35, United States Code, §120 of any United States application(s) listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior United States application in the manner provided by the first paragraph of Title 35, United States Code, §112, I acknowledge the duty to disclose material information as defined in Title 37, Code of Federal Regulations, §1.56(a) which occurred between the filing date of the prior application and the national or PCT international filing date of this application.



Docket Number: \_\_\_\_\_

(Application Serial No.)

**PCT/FR00/00822**

(Filing Date)

**March 31, 2000**

~~Patented, pending, abandoned~~

(Application Serial No.)

(Filing Date)

(Status - patented, pending, abandoned)

(Application Serial No.)

(Filing Date)

(Status - patented, pending, abandoned)

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

I hereby appoint the following person registered to practice before the Patent and Trademark Office as my attorney with full power of substitution and revocation to prosecute this application and all divisions and continuations thereof and to transact all business in the Patent and Trademark Office connected therewith and request that all correspondence be sent to him at the mailing address hereafter given:

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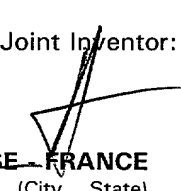
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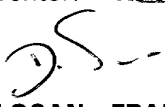
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